REMARKS

In the claim listing above, claims 91 and 99 have been amended. No claims have been added or canceled by this paper. Accordingly, as amended above, claims 99-103 are presented for further examination in this application.

Claim Amendments

As just indicated, claims 91 and 99 have been amended in the claim listing above.

In the process defined by claim 91, two steps, (c) and (d), have been amended. Step (c) now recites (c) "allowing said mixture to react under isostatic conditions of temperature, buffer and ionic strength to produce at least one *complementary* copy of said specific nucleic acid" . . . Step (d) in its amended form recites "removing **the** primer *portion* from the *complementary copy* produced in step (c) to regenerate a primer binding site on said specific nucleic acid, thereby allowing a new priming event to occur and producing more than one copy of said specific nucleic acid."

In a similar but not identical fashion, claim 99 has been amended in three ways. First, step (b)(ii) has been amended at the end by inserting the phrase *from a segment of said primer or primers*. Thus, this step now reads contacting . . . "(ii) one or more sequence unmodified primers each of which primer comprises at least one non-complementary sequence to a distinct sequence of said specific nucleic acid, such that upon hybridization to said specific nucleic acid at least one loop structure is formed *from a segment of said primer or primers*. Step (c) in claim 99 now recites at the end "thereby producing at least one *complementary* copy of said specific nucleic acid." Thirdly, step (d) has been amended to recite "removing *the loop structure or structures* from the *complementary* copy produced in step (c) to regenerate a primer binding site on said specific nucleic acid, to allow a previously presented priming event to occur and produce more than one copy of said specific nucleic acid.

Support for the foregoing amendments is drawn from the specification. See US 2008/0026372 A9 (January 31, 2008 – Corrected Publication), for example, **[0017]**, **[0018]** and **[0019]**. See also **[0055]**, **[0061]** and **[0062]**.

It is believed, therefore, that the foregoing amendments constitute subject matter to which they are duly entitled to claim.

Entry of the above amendments and new claim listing is respectfully requested.

Priority and Withdrawn Objections and Rejections

Applicants acknowledge with appreciation the indication in the Office Communication that their claim for priority (01/13/94) has been granted. They also appreciate the indication that the objections to the abstract, the specification and the claims have been mooted based upon amendments to the specification and the claims.

Commonality of Ownership

Applicants assert that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made.

The First Rejection Under 35 U.S.C. §103

Claims 91-95 and 98 stand rejected under 35 U.S.C. §103(a) as being unpatentable over Schuster et al. (US Patent 5,169,766 December 8, 1992) in view of Scheele (US Patent 5162209 November 10, 1992). According to the Office Communication (pages 4-6):

> Schuster et al. teaches a method of amplifying a nucleic acid molecule. With regard to Claim 91, Schuster et al. teaches providing a DNA target and mixing the target with nucleoside triphosphates (Figure 1 Column 7, lines 60-65). Schuster et al. teaches the use of primers which are chemically modified by blocking the 3' terminus using a 3' terminal nucleotide lacking a 3' hydroxyl group (Column 11 lines 60-64). Schuster

et al. teaches conditions or agents (nucleic acid producing catalysts) which increase amplification are present (Column 7, lines 50-55). Schuster et al. teaches that the assay mixture has a sufficient quantity of cofactors to support the degree of amplification desired (Column 7, lines 60-65). Schuster et al. teaches isostatic conditions, such as, the use of Tris base (pH stabilizer) in the amplification reactions, stable temperature of 37°C for 3 hours, and with a specific number of molecules of RNA (Column 13, lines 40-52). With regard to Claims 92 and 93, Schuster et al. teaches the use of RNase H to remove RNA from the cDNA (Column 8, lines 7-10). Schuster et al. teaches an mRNA promoter (primer) which is used to extend and make ssRNA (Figure

Schuster et al. teaches another primer (DNA) is annealed to the 55RNA and cDNA is copied (Figure 3). Schuster et al. teaches the ssRNA (which is the extended promoter) is destroyed by RNase H. Further, any primers which are in the solution but did not primer to the original ssDNA would be destroyed by Rnase H, therefore allowing for a reaction solution with only the cDNA that allows the completion of another cycle and the production of another cDNA strand identical to the ssDNA template. With regard to Claim 94, Schuster et al. teaches the primers can be DNA or RNA (Column 5 lines 35-38 and 55-60). With regard to Claim 95, Schuster et al. teaches the use of primers which are chemically modified by blocking the 3' terminus using a 3' terminal nucleotide lacking a 3' hydroxyl group (Column 11 lines 60-64). With regard to Claim 98, Schuster et al. teaches a promoter (primer) in which at least 1 nucleotide is noncomplementary (Fig 2 5th step).

However, though Schuster et al. teaches that the primer can be RNA (Column 11, lines 47-51), Schuster et al. does not teach that all of the primer sequences from the products are removed so that a primer binding site is regenerated on a specific nucleic acid allowing a new priming event to occur.

Scheele et al. teaches a method for preparing a dsDNA from an 55DNA (abstract). Scheele et al. teaches a method of providing a first DNA strand, contacting with a primer, synthesizing in the presence of the primer that contains RNA and producing a copy of the DNA template (abstract). Scheele et al. teaches that RNA primers can be digested using RNAse H (Column 6 lines 65-68). Therefore the RNA primers will be removed and the primer binding site is regenerated and as such all of the primer sequences from the products are removed so that a primer binding site is regenerated on a specific nucleic acid allowing a new priming event to occur.

Therefore it would be prima facie obvious to one of ordinary skill in the art to digest the RNA primer of Schuster et al. with the RNase H which

is already in the solution because Scheele et al. teaches that RNAse H digest RNA primers on a DNA/RNA hybrid strand. The ordinary artisan would be motivated to digest all RNA which is in the system in the double stranded form (e.g. DNA/RNA) in order to produce multiple copies of the nucleic acids of interest. Scheele et al. teaches that RNA primers can be digested using RNA5e H (Column 6 lines 65-68). Therefore the RNA primers will be removed and the primer binding site is regenerated.and as such all of the primer sequences from the products are removed so that a primer binding site is regenerated on a specific nucleic acid allowing a new priming event to occur.

The first obviousness rejection is respectfully traversed.

Applicants respectfully disagree with the characterization of Schuster's patent for several reasons. First, as now amended, the present invention calls for step (d) of removing the primer portion (claim 91) or the loop structure or structures (claim 99) from the complementary copy of the specific nucleic acid produced in step (c). The present invention and claims make it abundantly clear that the physical entity that is being removed in step (d) is the primer portion or the loop structure(s) from the previously produced complementary copy. In sharp contrast, Schuster et al. use RNase H to remove RNA transcripts from the template and not from the cDNA. Thus, Schuster et al. are describing the removal of RNA from an RNA transcript synthesized from a promoter. Schuster et al. do not describe removing the primer portion or loop structures *from the complementary copy*, as set forth in the present claims.

Second, the characterization that Schuster et al. teaches an mRNA promoter (primer) which is used to extend and make ssRNA (Figure 3) necessarily implies that a promoter and primer are synonymous or equivalent. This is not the case, however, because a promoter codes for primer-independent synthesis of an RNA transcript and a primer synthesizes new sequences by extension of the primer itself.

Third, the characterization that "any primers which are in the solution but did not primer to the original ssDNA would be destroyed by RNase H . . ." is incorrect because primers that are in solution are not substrates for RNase H.

Adding Scheele et al. to Schuster et al. would not have rendered Applicants' present invention and claims obvious to a person of ordinary skill in the art. The characterization in the rejection at hand that Scheele et al. teaches that RNA primers can be digested using RNase H (Column 6, lines 65-68) and that the RNA primers will be removed and the primer binding site is regenerated . . . clearly does not follow from the rest of the paragraph quoted in the rejection:

> . . . If either the primer or the tail is DNA, it will thereupon be rendered single-stranded, and can conveniently be removed using an appropriate ss DNA nuclease e.g. T4 polymerase will remove a 3' ss DNA overhang, as where the tail is DNA, while other nucleases (such as S1 nuclease, mung bean nuclease, and Exonuclease VII) are useful for removing a 5' ss DNA overhang, as where the primer is DNA. This removal of any ss DNA overhang left after removal of the RNA primer or tail results in a blunt-ended, full-length ds RNA suitable for further experimentation, including cloning.

Thus, Scheele et al. are describing treatment by RNase H followed by treatment with a nuclease which converts the polymer to a double-stranded blunt-ended molecule. This description in no way provides for the instantly claimed regeneration of a primer binding site and a new priming event.

It is respectfully submitted, therefore, that from a reading of the combined disclosures of Schuster et al. and Scheele et al., the ordinarily skilled artisan would not have reached the present invention and claims.

In view of the foregoing remarks and the above amendments to the claims, Applicants respectfully request reconsideration and withdrawal of the first obviousness rejection.

The Second Rejection Under 35 U.S.C. §103

Claims 91-98 stand rejected under 35 U.S.C. §103(a) as being unpatentable over Kacian et al, (US Patent 5554516 September 10,1996) in view of Scheele (US Patent 5162209 November 10, 1992). According to the Office Communication (pages 7-10):

> Kacian et al. teaches a method of amplifying a target nucleic acid sequence (Abstract). With regard to Claim 91, Kacian et al. teaches a method of incubating a promoter-primer and a target sequence in DNA priming and nucleic acid synthesizing conditions (ribonucleotide triphosphates and deoxyribonucleotide triphosphates) for a period of time to many multiple copies of the target sequence (Column 10 lines 23-33). Kacian et al. teaches using a DNA polymerase (nucleic acid producing catalyst) (Column 10 line 59). Kacian et al. teaches that the reaction takes place under conditions that are substantially isothermal and include substantially constant ionic strength and pH, i.e. isostatic conditions (Column 10 lines 37-45).
>
> With regard to Claims 92-93, Kacian et al. teaches that generation of target sequence is done using Rnase H (Column 4 lines 65-67 and Column 5 lines 1-5). Kacian et al. teaches the promoter-primer may be altered with ribonucleotides (Column 9, line 15). Therefore Kacian et al. teaches a reaction in which Rnase H is in the presence of a RNA-DNA hybrid (DNA target with a promoter with ribonucleotides), it is inherent that the Rnase H will denature the ribonucleotide promoter and thereby release the DNA target from the promoter.
>
> With regard to Claim 94, Kacian et al. teaches the use of DNA as a primer sequence (Column 6 lines 18-25). Kacian etal. teaches that this sequence may have modifications such as dideoxynucleotide residues that have been modified such as phosphorothioates (chemically) (Column 9, lines 14-16).
>
> With regard to Claim 95, Kacian et al. teaches the 3' end of the promoter-primer may be modified (Column 7, line 6). With regard to Claim 96, Kacian et al. teaches that one modification can be the addition of a phosphorothioate (sulphur heteroatom) (Column 9 lines 17).
>
> With regard to Claim 97, Kacian et al. teaches that promoter-primer can include the addition of 32' dideoxynucleotide residues modified with phosphorothioates (Column 9 lines 15-17). With regard to Claim 98, Kacian etal. teaches a promoter primer which has at least one nucleotide that is noncomplementary (Figure 1).
>
> However, though Kacian et al. does not teach all of the primer sequences from the products are removed so that a primer binding site is

regenerated on a specific nucleic acid allowing a new priming event to occur.

Scheele et al. teaches a method for preparing a dsDNA from an 5sDNA (abstract). Scheele et al. teaches a method of providing a first DNA strand, contacting with a primer, synthesizing in the presence of the primer that contains RNA and producing a copy of the DNA template (abstract). Scheele et al. teaches that RNA primers can be digested using RNAse H (Column 6 lines 65-68). Therefore the RNA primers will be removed and the primer binding site is regenerated and as such all of the primer sequences from the products are removed so that a primer binding site is regenerated on a specific nucleic acid allowing a new priming event to occur.

Therefore it would be prima facie obvious to one of ordinary skill in the art to digest the RNA primer of Kacian et al. with the RNAse H which is already in the solution because Scheele et al. teaches that RNA5e H digest RNA primers on a DNA/RNA hybrid strand. The ordinary artisan would be motivated to digest all RNA which is in the system in the double stranded form (e.g. DNNRNA) in order to produce multiple copies of the nucleic acids of interest. Scheele et al. teaches that RNA primers can be digested using RNAse H (Column 6 lines 65-68). Therefore the RNA primers will be removed and the primer binding site is regenerated and as such all of the primer sequences from the products are removed so that a primer binding site is regenerated on a specific nucleic acid allowing a new priming event to occur.

The second obviousness rejection is respectfully traversed.

At the outset, Applicants note that in their amplification process, Kacian et al. use RNA transcription to produce multiple copies of a target sequence. In contrast, Applicants' present invention and claims provides processes for producing more than one complementary copy of a specific nucleic acid through the regeneration of primer binding sites and the occurrence of new priming events. As recognized in the Office Communication (page 9), Kacian et al. does not teach that all of the primer sequences from the products are removed so that a primer binding site is regenerated on a specific nucleic acid to allow a new priming event to occur.

As indicated earlier in the first obviousness rejection, Scheele et al. does not cure the deficiency of the primary reference (in this case, Kacian et al.) with respect to regenerating a primer binding site and allowing a new priming event to occur. Scheele et al. teaches that RNA primers can be digested using RNase H (Column 6, lines 65-68) and that the RNA primers will be removed and the primer binding site is regenerated. But as quoted above in the first obviousness rejection, Scheele et al. follow the RNase H treatment with a subsequent treatment using a nuclease:

> . . . If either the primer or the tail is DNA, it will thereupon be rendered single-stranded, and can conveniently be removed using an appropriate ss DNA nuclease e.g. T4 polymerase will remove a 3' ss DNA overhang, as where the tail is DNA, while other nucleases (such as S1 nuclease, mung bean nuclease, and Exonuclease VII) are useful for removing a 5' ss DNA overhang, as where the primer is DNA. This removal of any ss DNA overhang left after removal of the RNA primer or tail results in a blunt-ended, full-length ds RNA suitable for further experimentation, including cloning. [Scheele et al. US 5,162,209, Column 7, lines 2-13]

Thus, in treating with RNase H followed by treatment with a nuclease, Scheele et al. converts the polymer to a double-stranded blunt-ended molecule. Such a description and such a nucleic acid molecule does not provide for the instantly claimed regeneration of a primer binding site and a new priming event.

For the foregoing reasons, the present invention and claims would not have been obvious to a person of ordinary skill in the art having the combined disclosures of Kacian et al. in view of Scheele et al.

Reconsideration and withdrawal of the second obviousness rejection is respectfully requested.

The Third Rejection Under 35 U.S.C. §103

Claims 96-97 stand rejected under 35 U.S.C. §103(a) as being unpatentable over Schuster et al. (US Patent 5,169,766 December 8, 1992) in view of Scheele (US

Patent 5,162,209 November 10, 1992) as applied to Claims 91-95 and 98 in view of Skerra (Nucleic Acids Research 1992 Vol.20 p.3551). According to the Office Communication (pages 11-12):

> Schuster et al. and Scheele et al., however, do not teach primers modified by heteroatoms comprised of nitrogen or sulfur and chemically modified primers comprised of nucleoside triphosphates.
>
> Skerra teaches a method of using phosphorothioate primers in an amplification method (Abstract). With regard to Claims 96-97, Skerra teaches the modification of primers by the addition of a single phosphorothioate bond (heteroatom of sulfur) at the first 3' terminal internucleotide linkage during synthesis of the oligodexoynucleotide (p. 3552, 1st column last paragraph). Skerra teaches that the phosphorothiate bond is much less favored substrate to nuclease activity than the naturally occurring phosphodiester bond (P. 3552 1st column last sentence and 2nd column 1st sentence).
>
> Therefore it would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made to modify the method of Schuster et al. and Scheele, to use the phosphorothioate primers as taught by Skerra. The ordinary artisan would have been motivated to modify the method of Schuster et al and Scheele, because Skerra teaches the use of phosphorothiate primers would avoid the lower PCR yield and non-specific side products resulting from 3' terminal editing of the primer molecule by protecting the oligodeoxynucleotide from a 3' terminal exonuleoytic attack (p. 3553 2nd column last paragraph).

The third obviousness rejection is respectfully traversed.

As indicated earlier in the first obviousness rejection, the combination of Schuster et al. in view of Scheele et al. would not have rendered Applicants' invention and claims obvious to a person of ordinary skill in the art. It must logically follow that the addition of yet a tertiary reference (in this case, Skerra) must be insufficient to reach any dependent claims because the added reference fails to cure the deficiencies in the first two references with respect to both independent claims at hand (91 & 99).

In view of the foregoing remarks and earlier remarks in the first obviousness rejection, Applicants respectfully request reconsideration and withdrawal of the third obviousness rejection.

The Fourth Rejection Under 35 U.S.C. §103

Claims 99-103 stand rejected under 35 U.S.C. §103(a) as being unpatentable over Schuster et al. (US Patent 5,169,766 December 8, 1992) in view of Scheele (US Patent 5,162,209 November 10, 1992) as applied to Claims 91-95 and 98 in view of Cerretti et al. (US Patent 5,317,087 May 31, 1994). According to the Office Communication (pages 13-14):

> Schuster et al. and Scheele et al., however, does not teach primer hybridization in which at least one loop structure is formed.
>
> Cerretti et al. teaches that a library of cDNA can be prepared by using hairpin loop primers (Column 11 lines 10-26). Cerretti et al. teaches the mRNA primer is hybridized to a first cDNA strand (Column 11 lines 10-26). Cerretti et al. teaches that this results in a "hairpin" loop at the 3' end of the initial cDNA strand that serves as an integral primer for the second DNA strand (Column 11 lines 10-26). Cerretti et al. teaches that the second cDNA strand is synthesized using a DNA polymerase and the hairpin loop is cleaved to produce double stranded cDNA molecules (Column 11 lines 10-26).
>
> Therefore it would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made to modify the method of Schuster et al. and Scheele et al. to use the hairpin loop primer as taught by Cerretti et al. The ordinary artisan would have been motivated to modify the method of Schuster et al. and Scheele et al. because Cerretti et al. teaches a method of using hairpin loops to copy small cDNA fractions from a large cDNA template (Column 11 lines 10-26). The ordinary artisan would want to use hairpin loops as a way to prepare a library of double-stranded cDNA and would want to cleave the mRNA primer from the target cDNA in order to keep using the original long strand of cDNA. The ordinary artisan would therefore be able to produce multiple copies at multiple positions of the target cDNA strand by annealing a mRNA primer, copying a fragment of cDNA with a hairpin loop, removing the mRNA primer, and adding another mRNA primer somewhere else on the target DNA.

The fourth obviousness rejection is respectfully traversed.

As in the case of the third obviousness rejection, it must logically follow that the addition of yet a tertiary reference (in this case, Cerretti et al.) to the first two cited references must be insufficient to reach any dependent claims because the added reference clearly fails to cure the deficiencies in the first two references with respect to both independent claims at hand (91 & 99).

Accordingly, Applicants respectfully request reconsideration and withdrawal of the fourth obviousness rejection.

The Rejection For Double Patenting

Claims 91-103 stand provisionally rejected on the ground of nonstatutory obviousness-type double patenting as being unpatentable over claims 112-148 of copending U.S. Patent Application Serial No. 10/713,183. According to the Office Communication (pages 16-17):

> Although the conflicting claims are not identical, they are not patentably distinct from each other because Claim 91-103 of the instant application describes the same method steps as Claim 112-148 of application 10/713,183. Both applications are drawn to a method of producing copies of a specific nucleic acid by providing a nucleic acid sample, contacting it with unmodified nucleic acid precursors and modified RNA primers. Both applications use a catalyst. Both applications modify primers using hereoatoms comprising nitrogen or sulfur. Both applications claims are drawn to primers, which comprise about 1 to about 200 noncomplementary nucleotide or nucleotide analogs.
>
> This is a provisional obviousness-type double patenting rejection because the conflicting claims have not in fact been patented.

In response, Applicants are submitting herewith accompanying this paper as Exhibit A their Terminal Disclaimer To Obviate A Provisional Double Patenting Rejection Over A Pending "Reference" Application [Form PTO/SB/25(12-08)].

In view of the submission of their Terminal Disclaimer (Exhibit A), Applicants respectfully request reconsideration and withdrawal of the double patenting rejection.

Early and favorable action is respectfully requested.

* * * * * * * *

SUMMARY AND CONCLUSIONS

In the above claim listing for this application, only claims 91 and 99 have been amended. No claims have been added or canceled by this paper.

No claim fee is believed due for this paper because the same number of claims are presented as previously paid for claims. This paper is also accompanied by a Request For Extension Of Time (3 months), a Terminal Disclaimer (Exhibit A) and authorization for the fee therefor. No other fee or fees are believed due in connection with this paper or the accompanying extension request. In the event that any other fee(s) is/are due in connection with this filing, however, the Patent and Trademark Office is hereby authorized to charge the amount of any such fee(s) to Deposit Account No. 05-1135, or to credit any overpayment thereto.

If a telephone call would be helpful in the processing of this paper or this application, Applicants' undersigned attorney requests that he be contacted at the numbers below.

Respectfully submitted,

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Dean L. Engelhardt et al.
Serial No.: 10/718,391 (Filed November 19, 2003)
Exhibit A To Applicants' January 27, 2009 Amendment Under 37 C.F.R. §1.115
(In Reply To The August 21, 2008 Office Communication)

EXHIBIT A